

Fumigant Toxicity of Four Essential Oils Against Two Major Stored Date Fruit Insects and Microbial Load

W.K.M. El-Shafei* and L.M. Lewaa

Department of Date Palm Pests and Diseases, Central Laboratory for Date Palm,
Agricultural Research Center (ARC), Giza, Egypt.

*Email address of the corresponding author: waelkamal27@yahoo.com

Abstract

El-Shafei, W.K.M. and L.M. Lewaa. 2025. Fumigant Toxicity of Four Essential Oils Against Two Major Stored Date Fruit Insects and Microbial Load. Arab Journal of Plant Protection, 43(1):87-95. <https://doi.org/10.22268/AJPP-001289>

Dates are among the most important fruits with high nutritional value. These fruits are exposed to many pests that reduce their market value which include *Oryzaephilus surinamensis* (L.) (Coleoptera: Silvanidae), *Plodia interpunctella* (Hübner) (Lepidoptera: Pyralidae), in addition to a microbial load. This work aimed to investigate the effect of four crude essential oil fumigants: lavender (*Lavandula officinalis*), orange (*Citrus sinensis*), marjoram (*Majorana hortensis*), and eucalyptus (*Eucalyptus citratus*), as a safe alternative to manage these pests. Five concentrations of each oil (62.5 to 1000 mg/L of air) were evaluated at 3, 5, and 7 days after treatment. Results obtained indicated that the fumigant toxicity increased with the increase of oils concentration and exposure time. *L. officinalis* essential oil had the highest toxic effect against the 4th larval stage of *P. interpunctella* and the *O. surinamensis* adults followed by *C. sinensis* which was much more toxic as fumigant than *M. hortensis* oil, whereas *E. citratus* oil was the least effective on the two tested insects. Results obtained also indicated clearly that Surinam adults were more tolerant to the four tested essential oils than the *Plodia* larvae. The four tested essential oils had high antibacterial and antifungal activity in the treated stored date fruits. This work clearly confirmed that lavender, orange, marjoram and eucalyptus oils can be used to protect stored date fruits due to their ability to control *P. interpunctella* and *O. surinamensis* insects as well as their ability to act as antibacterial and antifungal agents on stored date fruits.

Keywords: *Phoenix dactylifera*, essential oils, *Plodia interpunctella*, *Oryzaephilus surinamensis*, date fruits, microbial load.

Introduction

One of the most important fruits with high nutritional value is date palm fruit. The economy of some Arab countries in the Middle East and North Africa depends on its production and export (El-Shafei, 2011; El-Lakwah *et al.*, 2011; Johnson, 2012). Egypt produces around 1.7 million tons (FAOSTAT, 2020). The dry and semi-dry dates producing countries suffer from the infestation of stored dates by many insects, diseases and animal pests, which cause quantitative and qualitative loss and reduced market value (Mahmoud *et al.* 2022; Talukder *et al.* 2004). The Surinam beetle, *O. surinamensis* is considered a universal pest which attacks stored food products (El-Shafei, 2018; 2020; Panagiotakopulu & Buckland, 2017). Its larvae and adults can contaminate packaged foods, and consequently cause significant damage (Mowery *et al.*, 2002).

In addition, *P. interpunctella* (Plodia moth) is a widely spread and economically important pest of stored products (Rees, 2004; El-Shafei *et al.*, 2018; El-Shafei, 2020). Its larvae feed on broken grains, especially those that have been milled into goods like flour, preserved cereal products, dried vegetables and fruits, stored dates, meals, and processed foods (Veena *et al.*, 2005; Zinhoum *et al.*, 2019). Generally, synthetic pesticides (organophosphates and pyrethroids) and fumigants (phosphine) are used to manage this insect pest in storage systems (Kim *et al.*, 2014; Mbata & Shapiro-Ilana, 2010). Application of insecticides caused an increase in chemical residues in treated food products, the emergence of resistance in some insect populations, and human exposure

to pesticides (Arthur & Phillips, 2003; Phillips & Throne, 2010). Due to the chemical fumigants' negative environmental consequences and high cost, alternative control methods have been sought after. Plant essential oils can provide potential alternatives to control stored-product pests. Since the 1980s, numerous studies have concentrated on the fumigant toxicity of plant-derived essential oils and their constituents against insect pests of stored goods (Isman, 2006). A potential approach to preserve goods kept in warehouses is by using essential oils as a fumigant or contact agent against storage pests (Daferera *et al.*, 2000; Isman, 2000).

In order to evaluate the efficacy of four essential oils as fumigants, the current investigation was carried out for the control of two major insect pests, *P. interpunctella* and *O. surinamensis* and the microbial load of stored date fruits.

Materials and Methods

Tested insects and essential oils used

Date fruit insect species used in the current work included the Plodia moth, *P. interpunctella* and the Surinam beetle, *O. surinamensis* which were taken from the date pest mass rearing laboratory located in the Agricultural Research Center, Central Laboratory of Date Palm, department of date pests and diseases, Giza, Egypt.

The four tested oils (Table 1) were obtained from the Natural Oil Extraction Unit, National Research Center, Dokki, Giza, Egypt. These oils were tested against *P. interpunctella* larvae and *O. surinamensis* adults.

Toxicity of tested essential oil fumigants

To find out the fumigant toxicity of the four essential oils tested (*L. officinalis*, *C. sinensis*, *M. hortensis* and *E. citratus*) on *P. interpunctella* larvae and *O. surinamensis* adults, initially, a preliminary experiment was carried out to find out the effective concentrations of the four essential oils on the tested insects, which gave death rates from 20 to 80%. Based on this initial experiment, 62.5, 125, 250, 500 and 1000 ml/L of air concentrations were determined for fumigant toxicity against *Plodia* larvae and Surinam adults. Under laboratory conditions, these studies were conducted at a temperature of $27\pm 2^\circ\text{C}$ and a relative humidity of $65\pm 5\%$, according to the method described by EL-Shafei (2015), with some modifications.

The toxicity of these oil fumigants diluted with petroleum ether (40:60) was assessed on *P. interpunctella* larvae and *O. surinamensis* adults. In this experiment, 1000 ml plastic jars with tight lids served as the plant oil's fumigation chambers. Each oil was evaluated at doses of 62.5, 125, 250, 500 and 1000 mg/L of air inside the jars. In each treatment, five jars were used. One Whatman No. 1 filter paper strip, measuring 2×2 cm, was placed at the bottom of each jar. To achieve the specified oil dosage inside the tightly closed jars, 5 ml from each oil concentration was added to each plastic jar on filter paper. Thirty *P. interpunctella* larvae and *O. surinamensis* adults were placed separately in each jar in 10×15 cm gauze bags filled with approximately 50 g Siwi cultivar semi-dry dates and secured with rubber bands. At $27\pm 2^\circ\text{C}$ and $65\pm 5\%$ R.H., the jars were tightly sealed. In the control treatment, only petroleum ether was used. Mortality rates were determined 3, 5 and 7 days after treatment. The experiment was replicated four times.

Table 1. The main essential oils used in this study.

Scientific name	Family	Common name	Used plant part
<i>Majorana hortensis</i>	Lamiaceae	Marjoram	Flowers and leaves
<i>Lavandula officinalis</i>	Lamiaceae	Lavender	Leaves, flowers and spikes
<i>Citrus sinensis</i>	Rutaceae	Orange	Orange peel
<i>Eucalyptus citratus</i>	Myrtaceae	Eucalyptus	Leaves

Total count of bacteria, yeasts and molds

Total count of bacteria, yeasts and molds were carried out according to Assous *et al.* (2022) as follows: To each 10 grams of date fruit (Siwi cultivar) treated with essential oil, a 90 ml sterile saline solution (0.85% NaCl) was added and shaken well for 5 min. (to prepare 10^{-1} dilution) and the mixture was left for 15 min., and then serial dilutions up to sixth dilution were made. For the total bacterial count, 1 ml of each dilution of each treatment was poured into a sterilized petri dish and about 15 ml of plate count agar medium was poured in it and the petri dishes were then homogenized gently and left to solidify and then incubated at 37°C for 24-36 hrs. For yeasts and fungi counts, 1 ml of each dilution of each oil was added to a petri dish containing 15 ml of potato

dextrose agar (PDA) and incubated at 25°C for 5 days. The effect of treatments on bacteria, yeasts and fungi were estimated as log reduction according to Abd-Allah (2018).

Statistical analysis

Corrected mortality rates of tested insects treated with the four essential oils were determined according to Abbotts formula. The LC_{50} and LC_{90} values were determined using the probit analysis according to Finney, 1971. Statistical analysis to evaluate the mortality rate and microbial load data was conducted using SAS software (SAS Institute, 2006).

Results

Toxicity of four essential oil fumigants against *Plodia interpunctella* larvae

The efficacy of fumigant toxicity of five concentrations (62.5, 125, 250, 500 and 1000 mg/L of air) of four essential oils (*Lavandula officinalis*, *Citrus sinensis*, *Majorana hortensis* and *Eucalyptus citratus*) on *P. interpunctella* larvae 3, 5 and 7 days after exposure under laboratory conditions ($27\pm 2^\circ\text{C}$ and $65\pm 5\%$ R.H.) are summarized in Table 2. The findings showed that the *P. interpunctella* larvae's corrected mortality rates three days after exposure to the four tested oils mentioned above reached 32.50, 23.33, 16.67 and 3.83%, respectively, for the lowest concentration of 62.5 mg/L of air, and gradually increased by increasing the oils concentration to reach 62.50, 54.17, 46.67 and 34.17 % for the highest concentration of 1000 mg/L of air. Whereas the *P. interpunctella* larvae mortality rates 5 days after exposure to the four tested oils reached 55.83, 46.67, 40.00 and 29.17% for the lowest concentration of 62.5 mg/L of air and increased gradually with increased concentration to reach 85.00, 77.50, 70.00 and 57.50% at the highest concentration of 1000 mg/L of air. Seven days after treatment with the four tested oils, the mortality rates of *P. interpunctella* larvae reached 79.17, 70.00, 63.33 and 52.50% for the lowest concentration of 62.5 mg/L of air and reached 100.00, 94.17, 90.83 and 80.83% at the highest concentration of 1000 mg/L of air, for the four tested oils respectively.

Toxicity of four essential oils fumigants against *O. surinamensis* adults

The efficacy of fumigant toxicity of five concentrations (62.5, 125, 250, 500, and 1000 mg/L of air) of four essential oils (*Lavandula officinalis*, *Citrus sinensis*, *Majorana hortensis* and *Eucalyptus citratus*) on *O. surinamensis* adults under laboratory condition ($27\pm 1^\circ\text{C}$ and $65\pm 5\%$ R.H.) and different exposure periods of 3, 5 and 7 days after treatment are presented in Table 2. Results obtained indicated that the corrected mortality rate (%) of *O. surinamensis* adults three days after exposure to the four tested oils mentioned above at the concentration of 62.5 mg/L of air reached 25.83, 17.50, 8.33 and 3.33%, respectively, and increased gradually by increasing oils concentration to reach 55.83, 47.50, 37.50 and 27.50% at the highest concentration of 1000 mg/L of air, respectively. Whereas the mortality rate of *O. surinamensis* adults 5 days after exposure to the four tested oils reached 29.17, 64.17, 55.00 and 50.00% for the lowest concentration of 62.5 mg/L of air and increased gradually by increasing oils

concentration to reach 79.17, 70.83, 60.83 and 50.83% at the highest concentration of 1000 mg/L of air of the four tested oils, respectively. However, 7 days after treatment with the four tested oils, the mortality rate of *O. surinamensis* adults reached 72.50, 64.17, 55.00 and 50.00% in response to the lowest concentration of 62.5 mg/L of air and reached 100.00, 91.67, 84.17 and 74.17% at the highest concentration of 1000 mg/L of air for the four tested oils, respectively.

Lethal concentrations (LC₅₀ and LC₉₀) and toxicity index of four essential oil fumigants against the 4th larval stage of *P. interpunctella* and the adults of *O. surinamensis*, 7 days after treatment

The LC₅₀ and LC₉₀ of the four essential oils (*Lavandula officinalis*, *Citrus sinensis*, *Majorana hortensis* and *Eucalyptus citratus*) on *P. interpunctella* larvae under laboratory condition (27±2°C and 65±5% R.H.) are presented in Table 3. Results obtained showed that 7 days after exposure of 4th instar larvae of *P. interpunctella*, the LC₅₀ values reached 10.93, 16.35, 40.55 and 59.87 mg/L of air for *Lavandula officinalis*, *Citrus sinensis*, *Majorana hortensis*, and *Eucalyptus citratus* essential oils, respectively. Whereas for the *O. surinamensis* adults, the LC₅₀ values 7 days after exposure reached 16.354, 28.20, 39.13 and 75.63 mg/L of air for the same essential oils, respectively (Table 4). The calculated LC₉₀ values for the 4th instar larvae of *P. interpunctella*, reached 184.24, 583.97, 908.71 and 1942.01 mg/L of air, respectively (Table 3), compared to 319.90, 669.42, 1135.39 and 3875.28 mg/L of air, respectively, for the *O. surinamensis* adult's treatment (Table 4). Results obtained revealed that the toxicity effect of the four tested essential oils against the 4th instar larval of

P. interpunctella and the adults of *O. surinamensis* 7 days after treatment were in descending order as follows: *Lavandula officinalis*, *Citrus sinensis*, *Majorana hortensis*, and *Eucalyptus citratus*, indicating that the *Lavandula officinalis* essential oil was the best followed by *Citrus sinensis* which was much more toxic as fumigant than *Majorana hortensis* oil, whereas *Eucalyptus citratus* was the least effective essential oil against the two tested insects.. The results obtained also showed clearly that *P. interpunctella* larvae were more susceptible to the four tested essential oils than the adults of *O. surinamensis*. The correlation coefficient values of regression lines for the 4th larval stage of *P. interpunctella* and the adults of *O. surinamensis* indicated high significant correlation between the essential oils concentrations and insects' mortality rate.

The lethal times (LT₅₀ and LT₉₀) of fumigants against the 4th larval stage of *P. interpunctella* and *O. surinamensis* adults

The LT₅₀ and LT₉₀ values of the four tested essential oils showed a strong fumigant activity against the 4th larval stage of *P. interpunctella* and the adults of *O. surinamensis*, and as the oils concentrations increased (Tables 5 and 6), and the time required to achieve 50% and 90% mortality (LT₅₀ and LT₉₀) decreased. The findings demonstrated that the time needed to achieve 50% mortality (LT₅₀) of the 4th larval stage of *P. interpunctella* at the concentration of 500 mg/L of air were 2.14, 3.01, 3.71 and 4.89 days after fumigation with *Lavandula officinalis*, *Citrus sinensis*, *Majorana hortensis*, and *Eucalyptus citratus* essential oils, respectively (Table 5).

Table 2. Toxicity (mean ± SE) of four essential oil fumigants against *Plodia interpunctella* (Hübner) larvae and *O. surinamensis* adult at 27°C temperature and 65±5% relative humidity.

Oils	Conc. mg/L of air	Corrected larval mortality rates after different exposure periods (days)						Mean	
		3		5		7			
		PI Larvae	OS Adult	PI Larvae	OS Adult	PI Larvae	OS Adult	PI Larvae	OS Adult
<i>Lavandula officinalis</i> (Lavender)	Control	1.67±0.96 d	1.67±0.96 d	1.67±0.96 d	1.67±0.96 d	1.67±0.96 d	1.67±0.96 d	68.67±4.15 A	62.86±4.55 A
	62.5	32.50±0.83 c	25.83±1.60 c	55.83±1.60 c	49.17±1.60 c	79.17±1.60 c	72.50±0.83 c		
	125	40.00±1.36 bc	33.33±1.36 bc	63.33±1.36 bc	56.67±1.36 bc	86.67±1.36 bc	80.00±1.36 bc		
	250	45.00±0.96 abc	38.33±0.96 abc	68.33±0.96 abc	61.67±2.15 abc	90.00±1.36 abc	85.00±2.15 bc		
	500	52.50±0.83 ab	45.83±1.60 ab	75.83±1.60 ab	69.17±0.83 ab	93.33±0.00 ab	90.00±1.36 ab		
	1000	62.50±1.60 a	55.83±1.60 a	85.00±2.15 a	79.17±2.10 a	100±0.00 a	100.00±0.00 a		
<i>Citrus sinensis</i> (Orange)	Control	1.67±0.96 d	1.67±0.96 d	1.67±0.96 d	1.67±0.96 d	1.67±0.96 d	1.67±0.96 d	60.61±4.52 AB	54.67±4.59 AB
	62.5	23.33±1.36 c	17.50±2.10 c	46.67±1.36 c	40.83±0.83 c	70.00±1.36 c	64.17±0.00c		
	125	30.83±2.10 bc	25.00±1.67 bc	54.17±0.83 bc	48.33±0.96 bc	77.50±0.83 bc	71.67±0.96 bc		
	250	36.67±1.36 bc	30.00±1.92 bc	60.00±1.36 bc	53.33±1.36 bc	83.33±1.36 ab	76.67±1.36 bc		
	500	44.17±2.10 ab	37.50±1.60 ab	67.50±1.60 ab	60.83±1.60 ab	89.17±0.83 ab	84.17±0.83 ab		
	1000	54.17±1.60 a	47.50±0.83 a	77.50±1.60 a	70.83±0.83 a	94.17±0.83 a	91.67±0.96 a		
<i>Majorana hortensis</i> (Marjoram)	Control	1.67±0.96 d	1.67±0.96 d	1.67±0.96 d	1.67±0.96 d	1.67±0.96 d	1.67±0.96 d	53.33±4.61 BC	45.50±4.59 BC
	62.5	16.67±1.36 c	8.33±0.96 cd	40.00±1.92 c	31.67±1.67 c	63.33±1.36 c	55.00±0.96 c		
	125	23.33±1.36 bc	15.83±1.60 bcd	46.67±1.36 bc	39.17±0.83 bc	70.00±1.36 c	62.50±1.60 bc		
	250	28.33±0.96 bc	20.83±1.60 bc	51.67 ±0.96 bc	44.17±1.60 bc	75.00±2.15 bc	67.50±1.60 bc		
	500	35.83±0.83 ab	28.33±0.96 ab	59.17±0.83 ab	51.67±1.67 ab	82.50±1.60 ab	75.00±0.96 ab		
	1000	46.67±1.36 a	37.50±0.83 ab	70.00±1.36 a	60.83±1.60 a	90.83±1.60 a	84.17±0.83 ab		
<i>Eucalyptus citratus</i> (Eucalyptus)	Control	1.67±0.96 d	1.67±0.96 c	1.67±0.96 d	1.67±0.96d	1.67±0.96 d	1.67±0.96 d	42.33±4.49 C	37.67±3.90 C
	62.5	5.83±0.83 cd	3.33±1.36 c	29.17±1.60 c	26.67±0.00 c	52.50±0.83 c	50.00±1.36 c		
	5.53	12.50±1.60 bcd	8.33±0.96 bc	35.83±0.83 bc	31.67±1.67 bc	59.17±0.83 bc	55.00±0.96 bc		
	250	17.50±1.60 bc	12.50±0.83 bc	40.83±0.83 bc	35.83±1.60 bc	64.17±1.60 bc	59.17±0.83 bc		
	500	25.00±0.96 ab	20.00±0.00 ab	48.33±1.67 ab	43.33±0.00 ab	71.67±0.96 ab	66.67±1.36 ab		
	1000	34.17±1.60 a	27.50±0.83 a	57.50±0.83 a	50.83±1.60 a	80.83±1.60 a	74.17±0.83 a		

Values followed by the same letters in the same column are not significantly different at P=0.05.

Table 3. Lethal concentrations (LC₅₀ and LC₉₀) and toxicity index of four tested essential oil fumigants against *Plodia interpunctella* larvae at 27±2°C and 65±5% R.H. 7 days after treatment.

Oils	LC ₅₀ (mg/L air)	LC ₉₀ (mg/L air)	Confidence limits (mg/L of air)				Toxicity index at LC ₅₀	Slope± SD	r
			LC ₅₀		LC ₉₀				
			Lower	Upper	Lower	Upper			
<i>Lavandula officinalis</i> (Lavender)	10.93	184.24	0.60	22.02	108.00	487.00	100.00	1.045±0.337	0.999
<i>Citrus sinensis</i> (Orange)	16.35	583.97	3.37	29.93	285.13	835.32	66.84	0.825±0.204	0.996
<i>Majorana hortensis</i> (Marjoram)	40.55	908.71	21.70	57.23	435.54	1406.07	26.95	0.949±0.196	0.950
<i>Eucalyptus citratus</i> (Eucalyptus)	59.87	1942.01	35.68	84.11	717.45	23842.13	18.25	0.848±0.191	0.993

Table 4. Lethal concentrations (LC₅₀ and LC₉₀) and toxicity index of four tested essential oil fumigants against *Oryzaephilus surinamensis* adults at 27°C with 65±5% R.H. 7 days after treatment.

Oils	LC ₅₀ (mg/L air)	LC ₉₀ (mg/L air)	Confidence limits (mg/L of air)				Toxicity index at LC ₅₀	Slope± SD	r
			LC ₅₀		LC ₉₀				
			Lower	Upper	Lower	Upper			
<i>Lavandula officinalis</i> (Lavender)	16.35	319.90	1.75	28.89	157.19	895.15	100.00	0.992±0.318	0.998
<i>Citrus sinensis</i> (Orange)	28.20	669.42	12.60	42.15	307.10	1235.45	57.97	0.982±0.201	0.997
<i>Majorana hortensis</i> (Marjoram)	39.13	1135.39	18.87	56.87	491.39	1828.21	41.78	0.876±0.194	0.967
<i>Eucalyptus citratus</i> (Eucalyptus)	75.63	3875.28	45.94	113.07	1062.33	56831.64	21.62	0.749±0.189	0.989

Table 5. Lethal time (LT₅₀ and LT₉₀) values estimated at 500 and 1000 mg/L of air for *Plodia interpunctella* larvae.

Oils	Oil conc. (mg/L of air)	LT ₅₀ (days)	LT ₉₀ (days)	Confidence limits (days)				Slope± SD	r
				LT ₅₀		LT ₉₀			
				Lower	Upper	Lower	Upper		
<i>Lavandula officinalis</i> (Lavender)	500	2.14	8.40	2.42	3.87	7.14	11.02	1.578±0.267	0.973
<i>Citrus sinensis</i> (Orange)		3.01	11.81	3.24	4.11	9.40	17.76	3.609±0.524	0.993
<i>Majorana hortensis</i> (Marjoram)		3.71	14.46	1.66	5.67	8.95	36.18	1.579±0.266	0.964
<i>Eucalyptus citratus</i> (Eucalyptus)		4.89	19.50	4.40	5.45	11.41	54.82	3.346±0.515	0.994
<i>Lavandula officinalis</i> (Lavender)	1000	1.43	7.42	1.01	1.81	6.39	9.51	1.584±0.263	0.953
<i>Citrus sinensis</i> (Orange)		2.00	9.23	1.52	2.49	6.24	18.96	1.547±0.261	0.962
<i>Majorana hortensis</i> (Marjoram)		3.27	9.80	2.75	4.66	8.07	13.79	3.560±0.539	0.980
<i>Eucalyptus citratus</i> (Eucalyptus)		4.08	13.45	3.60	5.53	8.44	32.54	3.371±0.513	0.987

In addition, the time needed to cause 50% mortality (LT₅₀) of the 4th larval stage of *P. interpunctella* for the concentration 1000 mg/L of air were 1.43, 2.00, 3.27 and 4.08 days for the same essential oils, respectively (Table 5). Whereas the time needed to reach 50% mortality (LT₅₀) of *O. surinamensis* adults for the concentration 500 mg/L of air were 2.81, 3.88, 4.58 and 5.38 days after fumigation with the four essential oils, respectively (Table 6). Meanwhile, the time needed to obtain 50% mortality (LT₅₀) of *O. surinamensis* adults for the concentration 1000 mg/L air

were 1.86, 2.63, 3.83 and 4.65 days for the same essential oils, respectively (Table 6). The same trend was obtained for the LT₉₀ values for the tested insects. The correlation coefficient values of regression lines for the 4th larval stage of *P. interpunctella* and the adults of *O. surinamensis* indicated high significant correlation between the time of exposure to the four tested essential oils and insects' mortality rates. Results obtained indicated clearly that *O. surinamensis* adults were more tolerant of the four tested essential oils than the of *P. interpunctella* larvae.

Table 6. Lethal time values (LT₅₀ and LT₉₀) estimated at 500 and 1000 mg/L of air for *Oryzaephilus surinamensis* adults.

Oils	Oil conc. (mg/L of air)	LT ₅₀ (days)	LT ₉₀ (days)	Confidence limits (days)				Slope± SD	r
				LT ₅₀		LT ₉₀			
				Lower	Upper	Lower	Upper		
<i>Lavandula officinalis</i> (Lavender)	500	2.81	10.11	2.26	3.59	8.94	16.39	1.567±0.265	0.972
<i>Citrus sinensis</i> (Orange)		3.88	12.70	3.14	5.20	10.01	19.45	1.684±0.279	0.981
<i>Majorana hortensis</i> (Marjoram)		4.58	18.51	4.10	5.09	10.93	41.11	3.329±0.512	0.992
<i>Eucalyptus citratus</i> (Eucalyptus)		5.38	22.37	4.87	6.06	12.95	43.50	3.435±0.525	0.996
<i>Lavandula officinalis</i> (Lavender)	1000	1.86	9.45	1.40	2.32	7.95	19.05	1.554±0.260	0.959
<i>Citrus sinensis</i> (Orange)		2.63	10.56	2.09	3.33	8.45	17.68	1.554±0.264	0.969
<i>Majorana hortensis</i> (Marjoram)		3.83	13.03	3.34	4.25	7.55	19.29	3.345±0.518	0.985
<i>Eucalyptus citratus</i> (Eucalyptus)		4.65	15.29	4.17	5.17	9.06	22.75	3.229±0.513	0.993

Effect of four essential oils on total bacterial count of tested stored date fruits

The effect of different essential oils on total bacterial count on date fruit (Siwi cultivar) are summarized in Table 7. When the *Lavandula officinalis* oil was used, the mean total bacterial count mean for the concentration of 62.5 mg/L of air was 5.6×10^3 colony forming unit (cfu) with 1.6 log reduction compared with the control, and reached 2×10^3 cfu with log reduction 2.08 for the concentration 125 mg/L of air compared with the control. Whereas using oil concentrations of 250, 500 and 1000 mg/L of air, the total bacterial count was nil. As for the effect of *Citrus sinensis* oil, data showed that the total account was 2.6×10^4 cfu with 0.97 log reduction for the concentration of 62.5 mg/L of air, and 2.14×10^3 cfu with 2.06 log reduction for the concentration 125 mg/L of air compared with the control, and 1.64×10^3 with 2.18 log reduction for the concentration of 250 mg/L of air compared with the control. As for the concentrations of 500 and 1000 mg/L of air there were no bacterial growth. Results obtained from using *Majorana hortensis* oil showed that total bacteria count was 1.3×10^3 cfu with 2.26 log reduction for the concentration 62.5 mg/L of air compared with the control, and was 6.8×10^2 cfu with 2.55 log reduction for the concentration of 125 mg/L of air compared with the control, whereas there was no count at concentrations of 250, 500 and 1000 mg/L of air. Furthermore, when *Eucalyptus citratus* oil was used, the total bacterial count was 6.6×10^4 cfu with 0.56 log reduction for the concentration of 62.5 mg/L of air compared with the control. As for the concentration of 125mg/L of air, the total bacterial count was 2.1×10^4 with 1.06 log reduction compared with the control, and reached 1.7×10^3 cfu for the concentration of 250mg/L of air with 2.15 log reduction compared with the control and 1.3×10^3 cfu with log reduction 2.25 for the concentration of 500 mg/L of air compared with the control. The total mean count was nil when the oil concentration of 1000 mg/L of air was used.

Effect of four essential oils on total yeasts and molds count of tested stored date fruits

Total mean count of yeasts and molds in the treated date fruit (Siwi cultivar) with *Lavandula officinalis* oil was 1.26×10^3 cfu with 1.03 log reduction when the concentration of 62.5 mg/L of air was used compared to the control. The effect increased with an increase in concentration until no

microbial growth was observed when the concentration 1000 mg/L of air was used. Similar trend was obtained for the *Citrus sinensis* oil treatment. However, in the case of *Majorana hortensis* oil treatment, results showed that no microbial growth was observed when the concentrations 125, 250, 500 and 1000 mg/L of air were used. Furthermore, in the case of *Eucalyptus citratus* oil treatment, no microbial growth was observed when concentrations 500 and 1000 mg/L of air were used (Table 7).

Discussion

The current study showed that the four tested essential oils from *Lavandula officinalis*, *Citrus sinensis*, *Majorana hortensis*, and *Eucalyptus citratus* have a lethal fumigant activity on *P. interpunctella* larvae and the *O. surinamensis* adults. In general, essential oils have recently gained attention as potential pest control agents (Dimetry, 2014), and they have a quick degradability, selectivity, low mammalian toxicity, and little effect on the environment (Cloyd *et al.*, 2004). Fumigation is the most popular method used to manage pests in stored products because it is efficient against the majority of species, enables the insecticide to quickly reach the bug inside the grain, food, date fruits, and leaves with little residues (Kedia *et al.*, 2015; Phillips & Throne, 2010). Essential oils components with high vapor pressures are more likely to volatilize and are generally more toxic. According to Guleria & Tikku (2009), essential oils have fewer risks to non-target organisms due to their tendency to degrade through the action of sunlight, moisture, and detoxification enzymes.

The current investigation showed that the four tested essential oils have a fumigant toxicity effect against the 4th instar larval of *P. interpunctella* and the adults of *O. surinamensis* in the following descending order: *Lavandula officinalis*, *Citrus sinensis*, *Majorana hortensis*, and *Eucalyptus citratus*. The results obtained clearly indicated that the fumigants' toxicity increased with the increase of oils concentration and exposure period. Larvae of *P. interpunctella* were more vulnerable to the four tested essential oils than the adults of *O. surinamensis*. The adults of *O. surinamensis* and the fourth larval stage of *P. interpunctella* showed a very significant correlation between the essential oil concentrations and insect mortality rate, which is in agreement with a previous report (Aref *et al.*,

2015). Furthermore, *Eucalyptus globulus* oils were among the most significant oils with fumigant activity against the Surinam beetle, *Oryzaephilus surinamensis*, which attacks dry dates in storage (El-Salam *et al.* 2019). In 2011, Jianhua *et al.* reported that the oil from *Artemisia argyi* plant (Asteraceae: Artemisia) had a potent fumigant effect against *O. surinamensis*. Furthermore, Jesser *et al.* (2017) reported that six essential oils could be applied as possible biopesticides for controlling *P. interpunctella*.

The present study confirmed the effect of four tested essential oils on microbial load (bacteria, yeasts and molds) of stored date fruits showed high antibacterial and antifungal activity, which is agreement with previous findings (Kwiatkowski *et al.*, 2019; Rostami *et al.*, 2012; Todorova *et al.*, 2023).

The highest antimicrobial activity of Citrus essential oil in this study was at the concentration of 500 mg/L of air against bacteria and 1000 mg/L of air against yeasts and molds, which is in agreement with a previous study (Ngan *et al.*, 2020).

Majorana hortensis oil has many antimicrobial components that make this oil as one of the most significant essential oils

as an antimicrobial. Results obtained in this study revealed that this oil's impact was very effective at the concentration of 250 mg/L of air against bacteria and at the concentration of 125 mg/L of air against molds and yeasts, which is in agreement with a previous report (Gharib *et al.*, 2013). Eskandari *et al.* (2017) compared the antibacterial activity of essential oils of *Majorana hortensis*, *Trachyspermum copticum*, *Ducrosia anethifolia*, *Pulicaria gnaphalodes*, and *Foeniculum vulgare* against *Bacillus* sp. and *Listeria Monocytogenes* and concluded that the antibacterial efficacy of *Majorana hortensis*, *Trachyspermum copticum* and *Foeniculum vulgare* essential oils were the best.

Results of this study indicated that the essential oil of *Eucalyptus citratus* antimicrobial activity was low as compared with the other tested oils, which is in agreement with previous studies (Abdel-Fattah *et al.*, 2008; Cuéllar & Yunus, 2009). Mangalagiri *et al.* (2021) demonstrated that using Eucalyptus essential oil was the best antimicrobial agent against pathogenic bacteria and fungi which attack stored rice.

Table 7. The toxicity effect of four essential oil fumigants on the microbial load of stored date fruits.

Microbial load	Oils Conc. (mg/L air)	<i>Lavandula officinalis</i>			<i>Citrus sinensis</i>			<i>Majorana hortensis</i>			<i>Eucalyptus citratus</i>		
		log			log			log			log		
		Cfu/g	Cfu	R	Cfu/g	Cfu	R	Cfu/g	Cfu	R	Cfu/g	Cfu	R
Total counts	Control	2.46×10 ⁵ a	5.39	0.00	2.46×10 ⁵ ±a	5.39	0.00	2.46×10 ⁵ a	5.39	0.00	2.46×10 ⁵ a	5.39	0.00
	62.5	5.63×10 ³ b	3.75	1.64	2.62×10 ⁴ b	4.42	0.97	1.34×10 ³ b	3.13	2.27	6.67×10 ⁴ b	4.82	0.57
	125	2.01×10 ³ b	3.30	2.09	2.14×10 ³ c	3.33	2.06	6.87×10 ² b	2.84	2.55	2.11×10 ⁴ c	4.32	1.07
	250	0.00 b	0.00	5.39	1.64×10 ³ c	3.21	2.18	0.00 b	0.00	5.39	1.74×10 ³ d	3.24	2.15
	500	0.00 b	0.00	5.39	0.00 c	0.00	5.39	0.00 b	0.00	5.39	1.36×10 ³ d	3.13	2.26
	1000	0.00 b	0.00	5.39	0.00 c	0.00	5.39	0.00 b	0.00	5.39	0.00 d	0.00	5.39
	Log R mean	3.32 ab			2.66 ab			3.50 a			1.91 b		
Yeast and mold	Control	1.37×10 ⁴ a	4.14	0.00	1.37×10 ⁴ a	4.14	0.00	1.37×10 ⁴ a	4.14	0.00	1.37×10 ⁴ a	4.14	0.00
	62.5	126×10 ³ b	3.10	1.04	1.03×10 ⁴ b	4.01	0.13	3.2×10 ² b	2.51	1.63	6.2×10 ² b	2.79	1.35
	125	1.13×10 ³ b	3.05	1.09	9.47×10 ² c	2.98	1.16	0.00 b	0.00	4.14	5.07×10 ² bc	2.70	1.43
	250	8.07×10 ² bc	2.91	1.23	5.37×10 ² c	2.73	1.41	0.00 b	0.00	4.14	3.57×10 ² bc	2.55	1.59
	500	3.2×10 ² cd	2.51	1.63	2.37×10 ² c	2.37	1.76	0.00 b	0.00	4.14	0.00 c	0.00	4.14
	1000	0.00 d	0.00	4.14	0.00 c	0.00	4.14	0.00 b	0.00	4.14	0.00 c	0.00	4.14
	Log R mean	1.52 b			1.43 b			3.03 a			2.11 ab		

Cfu= Colony forming unit, Log R= Log ratio of reduction.

المخلص

الشافعي، وإيل كمال محمد ولؤي لواء منصور. 2025. سمية تبخير أربعة زيوت عطرية تجاه اثنتين من الحشرات الرئيسية لثمار التمور المخزونة وحملها الميكروبي. مجلة وقاية النبات العربية، 43(1):87-95. <https://doi.org/10.22268/AJPP-001289>

يعدّ التمر من أهم الثمار ذات القيمة الغذائية العالية. تتعرض هذه الثمار للعديد من الآفات، سواء كانت حشرية أو مرضية، مما يقلل من قيمتها التسويقية كمّاً ونوعاً، ومن بينها، الآفات الحشرية مثل فراشة *Plodia interpunctella* (Lepidoptera: Pyralidae) وخنفساء *Oryzaephilus surinamensis* (Coleoptera: Silvanidae)، بالإضافة للحمل الميكروبي على ثمار التمر. هدف هذا العمل إلى التحقق من تأثير تبخير أربعة زيوت عطرية خام، هي زيت اللافندر (*Lavandula officinalis*)، زيت الكافور (*Eucalyptus citratus*)، زيت البردقوش (*Majorana hortensis*)، وزيت البرتقال (*Citrus sinensis*) كبديل آمن لمكافحة هذه الآفات.

تم اختبار خمسة تراكيز من كل زيت (62.5 إلى 1000 مغ/ليتر من الهواء) لمكافحة هذه الآفات بعد فترات مختلفة من المعاملة (3، 5 و 7 أيام). وجد أن سمية مادة التبخير ازدادت مع زيادة تركيز الزيوت ومدة التعريض. كما أشارت النتائج إلى أن زيت اللافندر العطري كان أشدها سمية ضد يرقات العمر الرابع والحشرات الكاملة لحشرة خنفساء سورينام، يليه زيت البرتقال، ثم زيت الورد، وكان زيت الكافور أقلها سمية على الحشرات المختبرة. أشارت النتائج التي تم الحصول عليها بوضوح إلى أن الحشرات الكاملة لخنفساء سورينام كانت أكثر تحملاً للزيوت العطرية الأربعة المختبرة مقارنةً بيرقات فراشة البلوديا. كما أوضحت النتائج المتعلقة بالحمل الميكروبي (البكتيريا، الفطريات) أنه كان للزيوت العطرية المختبرة تأثير مضاد للبكتيريا والفطريات على ثمار التمر المخزنة. أكد هذا العمل بوضوح إمكانية استخدام زيوت اللافندر، البرتقال، الورد، الكافور في وقاية التمور المخزنة وذلك لقدرتها على مكافحة حشرات خنفساء سورينام وفراشة البلوديا، وخصائصها كمضاد للبكتيريا والفطريات على ثمار التمر المخزنة.

كلمات مفتاحية: زيوت عطرية، ثمار نخيل البلح، *Oryzaephilus surinamensis*، *Plodia interpunctella*، *Phoenix dactylifera*.

عناوين الباحثين: وائل كمال محمد الشافعي* ولؤي لواء منصور، قسم آفات وأمراض النخيل، المعمل المركزي للنخيل، مركز البحوث الزراعية، جيزة، مصر. *البريد الإلكتروني للباحث المراسل: waelkamal27@yahoo.com

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Received: July 19, 2023; Accepted: January 4, 2024

تاريخ الاستلام: 2023/7/19؛ تاريخ الموافقة على النشر: 2024/1/4